

HOW TO USE FACSCalibur

- First turn on the FACSCalibur and then the computer.
- Open the Cell Quest program.
- Open Detectors, Threshold and Compensation.
- Make sure the Machine is under pressure.
- Now prime the machine with the arm on the side and no liquid at the **SIP** at **LOW** speed.
- 2x Prime (priming takes out the air from the tubing).
- Threshold should be on FSC around 52.
- Now get the appropriate Dot Plots and Histograms.
- One of the Dot Plots should be FSC vs SSC and make sure the cells are on the screen, if not then adjust it with detectors.
- Now go to Dot Plot or Histogram for appropriate fluorochrome and 1st adjust the negative control with appropriate detectors.
- If using more than one fluorochrome simultaneously then make sure to have single label controls and adjust the compensation in Dot Plot to compensate, so that they are not over lapping.
- Collect the samples in the proper folder.
- After finish collecting the data, clean the machine --- Run the bleach for 5 minutes and then put the arm on the side and run it for 1 minute. Now run the distilled water for 5 minutes and then put the arm on the side and run it for 1 minute. Leave the machine in standby.